



09993966-082702

Atty. Dkt. No. 014024-0280733

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

Applicant: Michael Rohan

Title: Human and Non-Human Primate Homologues of Nkd Protein, Nucleic Acid  
Sequences Encoding, and Uses Thereof

Appl. No. 09/993,966

Filing Date: November 27, 2001

Examiner: NYA

Art Unit: 1646

**RECEIVED**  
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**AMENDMENT IN RESPONSE TO NOTICE UNDER 37 CFR §§1.821-825**

Commissioner for Patents  
Washington, D.C. 20231

Sir:

In response to the Notice to File Missing Parts of Nonprovisional Application mailed March 27, 2002, please amend the application as follows:

**In the Specification:**

Please amend the specification as shown:

Please delete the paragraph [0172] and replace it with the following paragraph:

**[0172]** The entire gene including the 5' and was cloned by the 5' RACE protocol using as the cDNA template for RACE cDNA synthesized from the total RNA obtained by LCM from a colon cancer patient as described above (RNA prep ID# 100/sample 1b3521 sample Name UC-C2CA) according to the manufacturers protocol (Clontech SMART RACE cDNA Amplification Kit, K1811-1) which is described below:

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5' RACE protocol:  
cDNA template for RACE was synthesized from RNA isolated by LCM from a cancer patient (RNA prep ID#100/Sample ID352/Sample name UC-C2CA) according to manufacturer's protocol (Clontech SMART RACE cDNA Amplification Kit, K1811-1). Amplification was performed with Clontech Advantage GC-cDNA PCR kit (K1907-1) at 1M GC according to manufacturer's protocol, using cDNA template above, universal primer mix provided by manufacturer and a primer specific for human Nkd (CH308: CTTGCCGTTGTTGTCAAAGTC) (SEQ ID NO: 23). PCR was carried out for 30 cycles of 94°C, 0.5 min/58°C, 0.5 min/68°C 2 min, followed by 10 cycles of 94°C, 1 min/58°C, 1 min/68°C 2 min. A final round of extension was carried out at 72°C for 10 min. The PCR products were cloned into pCR-TOPO4 (Invitrogen) and transformed into E. coli. Bacterial colonies harboring the correct 5'RACE product were identified by PCR screening using nested primers (CH306: CCCAGCATGGGGAAACTTCA (SEQ ID NO: 24) and CH308: CTTGCCGTTGTTGTCAAAGTC) (SEQ ID NO: 23).

Please delete the paragraph [0205] and replace it with the following paragraph:

**[0205]** In order to confirm the involvement of the hNkd gene in certain cancers, particularly colon cancer, a colon cancer cell line, SW620, was treated with  $\beta$ -catenin RC/AS oligos. Specifically, SC620 cells were tested with the following antisense oligos, CHIR30-5AS 5'-ACTCAGCTTGGTTAGTGTGTCAGGC-3', (SEQ ID NO: 25) and the reverse control oligos (CHIR30-5RC 5'-CGGACTGTGTGATTGGTTCGACTCA-3') (SEQ ID NO: 26). This experiment is described as follows.

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**REMARKS**

Applicant believes that the present application is now in condition for allowance. Favorable reconsideration of the application as amended is respectfully requested.

The Examiner is invited to contact the undersigned by telephone if it is felt that a telephone interview would advance the prosecution of the present application.

Respectfully submitted,

Date August 27, 2002

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By   
Samir Elamrani

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Should additional fees be necessary in connection with the filing of this paper, or if a petition for extension of time is required for timely acceptance of same, the Commissioner is hereby authorized to charge Deposit Account No 03-3975 for any such fees; and applicant hereby petitions for any needed extension of time.

**MARKED UP VERSION ATTACHED TO AMENDMENT IN****SERIAL NO. 09/993,966****Marked up version of paragraph [0172] is below:**

**[0172]** The entire gene including the 5' and was cloned by the 5' RACE protocol using as the cDNA template for RACE cDNA synthesized from the total RNA obtained by LCM from a colon cancer patient as described above (RNA prep ID# 100/sample 1b3521 sample Name UC-C2CA) according to the manufacturers protocol (Clontech SMART RACE cDNA Amplification Kit, K1811-1) which is described below:

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